

What is claimed is:

1. A method for producing α -tocopherol or α -tocopheryl esters comprising:

- 5 (a) biologically producing a compound selected from the group consisting of farnesol and geranylgeraniol; and
- (b) chemically converting said compound into α -tocopherol or an α -tocopheryl ester.

2. The method of Claim 1, wherein said compound is geranylgeraniol having a first, second, third and fourth
10 olefin moieties and wherein said step of chemically converting comprises:

- (a) reducing at least one of said second, third and fourth olefin moieties of said geranylgeraniol to form an allylic alcohol; and
- 15 (b) forming α -tocopherol or an α -tocopheryl ester from said allylic alcohol and a hydroquinone.

3. The method of Claim 2, wherein said reducing step (a) comprises:

adding a protecting group to said geranylgeraniol to
20 protect the first olefin moiety of said geranylgeraniol from reduction; and

reducing at least one of the second, third and fourth olefin moieties of said protected geranylgeraniol; and

removing said protecting group from said reduced protected geranylgeraniol to provide said allylic alcohol.

4. The method of Claim 3, wherein said protecting group is a hydroxy protecting group.

5 5. The method of Claim 4, wherein said hydroxy protecting group is an ester.

6. The method of Claim 5, wherein said ester is selected from the group consisting of isobutyrate and pivaloate.

10 7. The method of Claim 2, wherein said reducing step comprises hydrogenation.

8. The method of Claim 2, wherein said allylic alcohol comprises phytol.

15 9. The method of Claim 2, wherein formation of said α -tocopherol or an α -tocopheryl ester comprises contacting said allylic alcohol with an acid in the presence of said hydroquinone.

10. The method of Claim 2, wherein said hydroquinone is a trimethyl hydroquinone.

20 11. The method of Claim 10, wherein said trimethyl hydroquinone is 2,3,5-trimethyl hydroquinone.

12. The method of Claim 1, wherein said compound is farnesol and wherein said step of chemically converting comprises:

(a) converting farnesol to a first intermediate comprising sufficient carbon atoms to form a trimethyltridecyl substituent group of vitamin E when said intermediate is reacted with a hydroquinone to form vitamin
5 E; and

(b) reacting said intermediate compound with a hydroquinone to form vitamin E.

13. The method of Claim 12, wherein said converting step comprises:

10 oxidizing said farnesol to farnesal; and
forming a methyl ketone from said farnesal.

14. The method of Claim 13, further comprising:
reducing olefin moieties in said methyl ketone to form
an alkyl methyl ketone; and

15 forming an allylic alcohol from said alkyl methyl
ketone.

15. The method of Claim 12, wherein said intermediate comprises isophytol.

16. The method of Claim 12, wherein said hydroquinone
20 is a trimethyl hydroquinone.

17. The method of Claim 16, wherein said trimethyl hydroquinone is 2,3,5-trimethyl hydroquinone.

18. The method of Claim 1, wherein said compound is farnesol and wherein said step of chemically converting
25 comprises:

- (a) oxidizing said farnesol to farnesal;
- (b) forming a methyl ketone from said farnesal;
- (c) reducing olefin moieties in said methyl ketone to form an alkyl methyl ketone;

5 (d) forming an allylic alcohol from said alkyl methyl ketone, wherein said allylic alcohol comprises a sufficient number of carbon atoms to form at least a trimethyltridecyl substituent group of vitamin E when vitamin E is formed from said allylic alcohol and a
10 corresponding hydroquinone; and

- (e) forming vitamin E from said compound and a hydroquinone.

19. The method of Claim 18, wherein said allylic alcohol comprises isophytol.

15 20. The method of Claim 18, wherein said hydroquinone is a trimethyl hydroquinone.

21. The method of Claim 20, wherein said trimethyl hydroquinone is 2,3,5-trimethyl hydroquinone.

20 22. The method of Claim 1, wherein said compound is farnesol.

23. The method of Claim 1, wherein said compound is geranylgeraniol.

24. A method for producing α -tocopherol or α -tocopheryl esters comprising:

(a) culturing a microorganism in a fermentation medium to produce a product selected from the group consisting of farnesyl phosphate and farnesol, wherein the action of squalene synthase of said microorganism is reduced;

(b) recovering said product; and

(c) chemically converting said product into α -tocopherol or an α -tocopheryl ester.

25. The method of Claim 24, wherein said fermentation medium comprises a squalene synthase inhibitor.

26. The method of Claim 24, wherein said microorganism is genetically modified to decrease the action of squalene synthase.

27. The method of Claim 26, wherein said microorganism is further genetically modified to increase the action of HMG-CoA reductase.

28. The method of Claim 27, wherein the action of HMG-CoA reductase is increased by overexpression of HMG-CoA reductase or the catalytic domain thereof in the microorganism.

29. The method of Claim 28, wherein said microorganism is further genetically modified to increase the action of a protein selected from the group consisting of acetoacetyl Co-A thiolase, HMG-CoA synthase, mevalonate kinase,

phosphomevalonate kinase, phosphomevalonate decarboxylase, isopentenyl pyrophosphate isomerase, farnesyl pyrophosphate synthase, D-1-deoxyxylulose 5-phosphate synthase, and 1-deoxy-D-xylulose 5-phosphate reductoisomerase.

5 30. The method of Claim 29, wherein the microorganism has been genetically modified to increase the action of farnesyl pyrophosphate synthase.

 31. The method of Claim 30, wherein the microorganism has been genetically modified to overexpress farnesyl
10 pyrophosphate synthase.

 32. The method of Claim 24, wherein said microorganism is an *erg9* mutant.

 33. The method of Claim 32, wherein said microorganism comprises a *erg9Δ::HIS3* deletion/insertion allele.

15 34. The method of Claim 24, wherein said recovering step comprises recovering said product from said microorganism.

 35. The method of Claim 24, wherein said product is secreted into said fermentation medium by said microorganism
20 and wherein said step of recovering comprises purification of said product from said fermentation medium.

 36. The method of Claim 24, wherein said product is intracellular farnesyl phosphate and farnesol and said step

of recovering comprises isolating said farnesyl phosphate and farnesol from said microorganism.

37. The method of Claim 24, wherein said product is intracellular farnesyl phosphate and said step of recovering
5 further comprises dephosphorylating said farnesyl phosphate to produce farnesol.

38. The method of Claim 24, wherein said microorganism is a fungi.

39. The method of Claim 38, wherein said fungi has
10 been genetically modified to express at least a portion of the enzymes in the mevalonate independent pathway.

40. The method of Claim 39, wherein said fungi has been genetically modified to express an enzyme selected from the group consisting of D-1-deoxyxylulose 5-phosphate
15 synthase, and 1-deoxy-D-xylulose 5-phosphate reductoisomerase.

41. The method of Claim 38, wherein said fungi is a yeast and said yeast is blocked in the ergosterol pathway and is genetically modified to take up exogenous sterols
20 under aerobic conditions.

42. The method of Claim 24, wherein said microorganism is a bacteria.

43. The method of Claim 42, wherein said bacteria has been genetically modified to express at least a portion of
25 the enzymes in the mevalonate pathway.

44. The method of Claim 43, wherein said bacteria has been genetically modified to express an enzyme selected from the group consisting of acetoacetyl Co-A thiolase, HMG-CoA synthase, HMG-CoA reductase, mevalonate kinase, phosphomevalonate kinase, and phosphomevalonate decarboxylase.

45. The method of Claim 24, wherein said microorganism is a microalgae.

46. The method of Claim 45, wherein said microalgae is selected from the group consisting of *Chlorella* and *Prototheca*.

47. The method of Claim 24, wherein said microorganism has been genetically modified to increase phosphatase action.

48. A method for producing α -tocopherol or α -tocopheryl esters comprising:

(a) culturing a microorganism in a fermentation medium to produce a product selected from the group consisting of geranylgeranyl phosphate and geranylgeraniol, wherein the
5 action of squalene synthase of said microorganism is reduced;

(b) recovering said product; and

(c) chemically converting said geranylgeraniol into α -
10 tocopherol or an α -tocopheryl ester.

49. The method of Claim 48, wherein said fermentation medium comprises a squalene synthase inhibitor.

50. The method of Claim 48, wherein said microorganism is genetically modified to decrease the action of squalene
15 synthase.

51. The method of Claim 50, wherein said microorganism is further genetically modified to increase the action of HMG-CoA reductase.

52. The method of Claim 48, wherein the action of HMG-
20 CoA reductase is increased by overexpression of HMG-CoA reductase or the catalytic domain thereof in the microorganism.

53. The method of Claim 52, wherein said microorganism is further genetically modified to increase the action of a
25 protein selected from the group consisting of acetoacetyl

Co-A thiolose, HMG-CoA synthase, mevalonate kinase,
phosphomevalonate kinase, phosphomevalonate decarboxylase,
isopentenyl pyrophosphate isomerase, farnesyl pyrophosphate
synthase, geranylgeranyl pyrophosphate synthase, D-1-
5 deoxyxylulose 5-phosphate synthase, and 1-deoxy-D-xylulose
5-phosphate reductoisomerase.

54. The method of Claim 53, wherein the microorganism
has been genetically modified to increase the action of
farnesyl pyrophosphate synthase.

10 55. The method of Claim 54, wherein the microorganism
has been genetically modified to overexpress farnesyl
pyrophosphate synthase.

56. The method of Claim 53, wherein the microorganism
has been genetically modified to increase the action of
15 geranylgeranyl pyrophosphate synthase.

57. The method of Claim 54, wherein the microorganism
has been genetically modified to overexpress geranylgeranyl
pyrophosphate synthase.

58. The method of Claim 48, wherein said microorganism
20 is an *erg9* mutant.

59. The method of Claim 58, wherein said microorganism
comprises a *erg9Δ::HIS3* deletion/insertion allele.

60. The method of Claim 48, wherein said recovering step comprises recovering said product from said microorganism.

5 61. The method of Claim 48, wherein said product is secreted into said fermentation medium by said microorganism and wherein said step of recovering comprises purification of said product from said fermentation medium.

10 62. The method of Claim 48, wherein said product is intracellular geranylgeranyl phosphate and geranylgeraniol and said step of recovering comprises isolating said geranylgeranyl phosphate and geranylgeraniol from said microorganism.

15 63. The method of Claim 48, wherein said product is intracellular geranylgeranyl phosphate and said step of recovering further comprises dephosphorylating said geranylgeranyl phosphate to produce geranylgeraniol.

64. The method of Claim 48, wherein said microorganism is a fungi.

20 65. The method of Claim 64, wherein said fungi has been genetically modified to express at least a portion of the enzymes in the mevalonate independent pathway.

66. The method of Claim 65, wherein said fungi has been genetically modified to express an enzyme selected from the group consisting of D-1-deoxyxylulose 5-phosphate

synthase, and 1-deoxy-D-xylulose 5-phosphate reductoisomerase.

67. The method of Claim 64, wherein said fungi is a yeast and said yeast is blocked in the ergosterol pathway and is genetically modified to take up exogenous sterols under aerobic conditions.

68. The method of Claim 48, wherein said microorganism is a bacteria.

69. The method of Claim 68, wherein said bacteria has been genetically modified to express at least a portion of the enzymes in the mevalonate pathway.

70. The method of Claim 69, wherein said bacteria has been genetically modified to express an enzyme selected from the group consisting of acetoacetyl Co-A thiolase, HMG-CoA synthase, HMG-CoA reductase, mevalonate kinase, phosphomevalonate kinase, and phosphomevalonate decarboxylase.

71. The method of Claim 48, wherein said microorganism is a microalgae.

72. The method of Claim 71, wherein said microalgae is selected from the group consisting of *Chlorella* and *Prototheca*.

73. The method of Claim 48, wherein said microorganism has been genetically modified to increase phosphatase action.

74. A method for producing α -tocopherol or α -tocopheryl esters comprising:

(a) biologically producing a first compound selected from the group consisting of geranylgeraniol and

5 geranylgeranyl pyrophosphate;

(b) contacting said first compound with a geranylgeranyl reductase to form a second compound selected from the group consisting of phytol and phytyl diphosphate; and

10 (c) chemically converting said second compound into α -tocopherol or an α -tocopheryl ester.

75. A method, as claimed in Claim 74, wherein said step of contacting is conducted *in vivo* using a microorganism having geranylgeranyl reductase activity.

15 76. A method, as claimed in Claim 74, wherein said step of contacting is conducted in a biotransformation process.

77. A method, as claimed in Claim 74, wherein said first compound is geranylgeraniol and said second compound
20 is phytol.

78. A method, as claimed in Claim 74, wherein said first compound is geranylgeranyl pyrophosphate and said second compound is phytyl diphosphate.

79. A method, as claimed in Claim 74, wherein said step of chemically converting comprises reacting said second compound or a derivative thereof with a hydroquinone to form α -tocopherol or an α -tocopheryl ester.

5 80. A method, as claimed in Claim 74, wherein said step of contacting comprises purifying said first compound and transforming said first compound with isolated geranylgeranyl reductase or microorganisms having geranylgeranyl reductase activity.

10 81. The method of Claim 80, wherein said microorganism is further genetically modified to increase the action of geranylgeranyl reductase.

15 82. The method of Claim 81, wherein the action of geranylgeranyl reductase is increased by overexpression of geranylgeranyl reductase.

83. A method for producing a compound selected from the group consisting of farnesol and geranylgeraniol comprising:

- (a) culturing a microorganism in a fermentation medium comprising a compound selected from the group consisting of isoprenol and prenol to produce a product selected from the group consisting of farnesyl phosphate, farnesol, geranylgeranyl phosphate and geranylgeraniol; and
- (b) recovering said product.

84. The method of Claim 83, wherein said microorganism is further genetically modified to increase the action of dimethylallyl transferase.

85. The method of Claim 84, wherein the action of dimethylallyl transferase is increased by overexpression of dimethylallyl transferase in the microorganism.

86. The method of Claim 85, wherein the microorganism has been genetically modified to increase the action of farnesyl pyrophosphate synthase.

87. The method of Claim 86, wherein the microorganism has been genetically modified to overexpress farnesyl pyrophosphate synthase.

88. The method of Claim 85, wherein the microorganism has been genetically modified to increase the action of geranylgeranyl pyrophosphate synthase.

89. The method of Claim 86, wherein the microorganism has been genetically modified to overexpress geranylgeranyl pyrophosphate synthase.

5 90. The method of Claim 83, wherein the microorganism has been genetically modified to increase the action of an enzyme selected from the group consisting of isoprenol kinase and prenol kinase.

10 91. The method of Claim 83, wherein the microorganism has been genetically modified to overexpress an enzyme selected from the group consisting of isoprenol kinase and prenol kinase.

92. The method of Claim 83, wherein said microorganism is an *erg9* mutant.